

## Final Report to National Aeronautics and Space Administration

For Grant No. NSG-78-60

## Part II. The Use of Pathogen-Free Plants in a Microcosm

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January 1, 1966-June 30, 1966

The level of support for "The Use of Pathogen-Free Plants in a Microcosm" was cut back and the studies on the "Effect of High Intensity Light in Plant Growth" received most of the funds. The small amount of funds left over for this part were used for the development and evaluation of a prototype germ-free, controlled environment, plant growth chamber. A report on this chamber is attached.

A paper is now being prepared on the effects of growing plants under gnotobiotic conditions. This paper will soon be submitted to Phytopathology for publication. ✓

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## INTRODUCTION

A prototype germfree, controlled environment, plant growth chamber was installed in the Gnotobiotic Laboratory of Colorado State University in September 1965 under National Science Foundation Grant GB-821. Prior to this time, some difficulty was experienced in securing the cooperation of the Physical Plant of Colorado State University for the construction of the unit; however, two senior students in electrical engineering were engaged and the first configuration was completed. Initial tests of this unit established the feasibility of the design but modifications were necessary. A full time graduate student with bioengineering background was engaged to continue work on design and modification. This report outlines progress in construction and modification of the prototype which is essentially completed; however, further improvements, also outlined in this report, are in the process.

## GENERAL DESCRIPTION

The main physical features of the unit, which were outlined in the original proposal to the National Science Foundation, are shown in Fig. 1A and 2A. The complete unit consists of two main chassis, each with several sub units.

The Air Handling Chassis (Fig. 1A, 1B) has two sub units, and air conditioning and a control sub unit. The air conditioning sub unit consists of five (5) 725 watt preheating elements, dehumidifying coils, cooling coils, five (5) 725 watt reheating elements, a spray humidifier, two (2) compressor-condensers for the coils, and a variable speed blower motor. The control sub unit contains the necessary electronic and electrical components to receive temperature and humidity sensor signals, convert, interpret and apply the necessary modulating commands to the aforementioned air conditioning group. The Air Handling Chassis should supply: 400-800 c.f.m. air volume at temperatures of 10°-35° centigrade with relative humidities of 10%-85%.

The Chamber Chassis (Fig. 2A, 2B) also has two sub units, the tent and the lighting sub unit. The tent sub unit has two chambers, one within the other. The outer shell is constructed of rigid, transparent, 1/4 inch acrylic plastic approximately 35 inches deep, 45 inches wide and 45 inches high which provides excellent visibility and a mounting surface for: the airlock, the supply and return ducts, the gloves and the lights. The inner shell, the germfree area, is constructed of flexible, transparent, 12 mil polyvinyl chloride approximately 35 inches deep, 45 inches wide and 40 inches high which is chemically stable, allows excellent visibility and is inexpensive to repair,

modify or replace. This inner shell or tent, operating at a positive pressure of approximately 3/8 inch water guage to compensate for small leaks and to maintain a negative contamination gradient, houses the stainless steel plant growth platform which is perforated to hold 48 clay pots and allow vertical airflow. This platform is mounted on a bearing attached to a hydraulic jack, thus, it can be raised or lowered manually from the outside and manually rotated from the inside. Normally, the platform is raised higher than the airlock and gloves to eliminate light imbalances caused by them and to also provide a suitable light intensity in conjunction with switches on the light bank. When it is necessary to attend to the plants, the platform is temporarily lowered to make the plants accessible to the airlock and the gloves. The lighting unit rests on the rigid shell and can be removed to reduce the weight and height of the Chamber Chassis for purposes of transportation. This unit consists of: a bank of twenty (20), 48 inch, 110 watt fluorescent lights with their ballasts: four (4), 100 watt incandescent lights, two (2) cooling motors and control switches. The switches are wired so that they can provide all the fluorescents or half the fluorescents, with or without the incandescents, thus, light quantity can be varied.

The germfree filter assemblies, also a part of the Chamber Chassis, are situated so as to continuously filter all of the supply air and all of the return air.

### THEORY OF OPERATION

All surfaces on the germfree or tent side of the air filters are a potential source of contamination, thus, sterilization of these surfaces is the first step in the operation of the unit. The chamber and the germfree filters are sealed off from the Air Handling Chassis and the external environment, a 2% solution of peracetic acid in water is then sprayed into the tent under pressure which perfuses fumes to all surfaces in contact with the filtered air. The fumes are retained in the sealed area for at least 1/2 hour and then exhausted to the atmosphere for approximately 4 hours after which time the Chamber Chassis is connected to the Air Handling Chassis and the unit is then ready for germfree operation.

The desired temperature and humidity conditions are preset at the control unit of the Air Handling Chassis and the chamber is allowed to come to equilibrium. At that time, the chamber is ready to receive germfree plants and apparatus from a transfer tent through the airlock. The plant growth platform is lowered to receive the plant pots which are inserted in the holes provided for them. Light quantity is a function of the height of the plant

growth platform which is normally raised above the level of the airlock as previously explained, in conjunction with the light switches found on the light bank. Day-night cycles are controlled by timers located at the laboratory power panel and accurate, permanent record monitoring of temperature and humidity levels is accomplished by a remote multichannel strip recorder. Nutrient solution can be introduced, whenever needed, from a transfer tent through the airlock while still retaining germfree conditions. Should the plants require more growth room than the 22 inches available with the plant growth platform at its normal growth height, the platform can be lowered for a maximum growth height of approximately 40 inches.

### MODIFICATIONS TO THE PROTOTYPE

After construction, a two stage program of evaluation and modification was initiated concomittant with the two-chassis configuration of the unit (Figs. 1A, 2A): a) the Air Handling Chassis to be evaluated and modified in its areas of air supply and air conditioning; b) the Chamber Chassis to be evaluated and modified with respect to its germfree abilities, air distribution and illumination. Figures 1A and 2A represent the original prototype and Figures 1B and 2B represent the prototype as presently modified.

#### Stage I- Completed Modifications of the Air Handling Chassis (Fig. 1B)

- a. There was considerable icing of the dehumidifying and cooling coils after 48 hours of operation. This was reduced by: re-locating the blower motor, streamlining the air shrouds and reducing the volume from 800 c.f. m. to 400 c.f. m. , the net effect was to reduce air layering over the coils.
- b. Air flow resistance was reduced by rotating the five preheating and the five reheating elements 90 degrees and perforating their mountings. Figure 3
- c. The preheating and reheating elements were rewired to provide more even heating of the air.
- d. The spray humidifier was relocated to reduce oxidation and mineralization of the reheating elements.
- e. The air supply plenum was altered from two 8 inch ducts to one 10 inch duct. Two reducing "Y's" (10 inch to 8 inch) were installed in both lines to feed the Chamber Chassis supply and return fittings.

- f. Fresh air mixture functions were removed from the return plenum located under the Chamber Chassis and assigned to each of the aforementioned reducing "Y's" located between the two main chassis.

Stage I- Modifications in Progress on the Air Handling Chassis include the following:

- a. The suction manifold of the dehumidifying and cooling coils are being modified to make these coils more reactive to an icing condition.
- b. The dehumidifying and cooling compressors are being converted from a cycling to a continuous mode of operation to reduce power consumption and maintenance.

Stage II- The Chamber Chassis germfree filter system is being improved to provide better than 99.95% filtration of particles larger than .3 microns. Improved means for venting peracetic acid fumes (used to sterilize the chamber) and protection of all surfaces that may come in contact with the fumigant are also under study.

#### TENTATIVE EVALUATION OF ORIGINAL UNIT

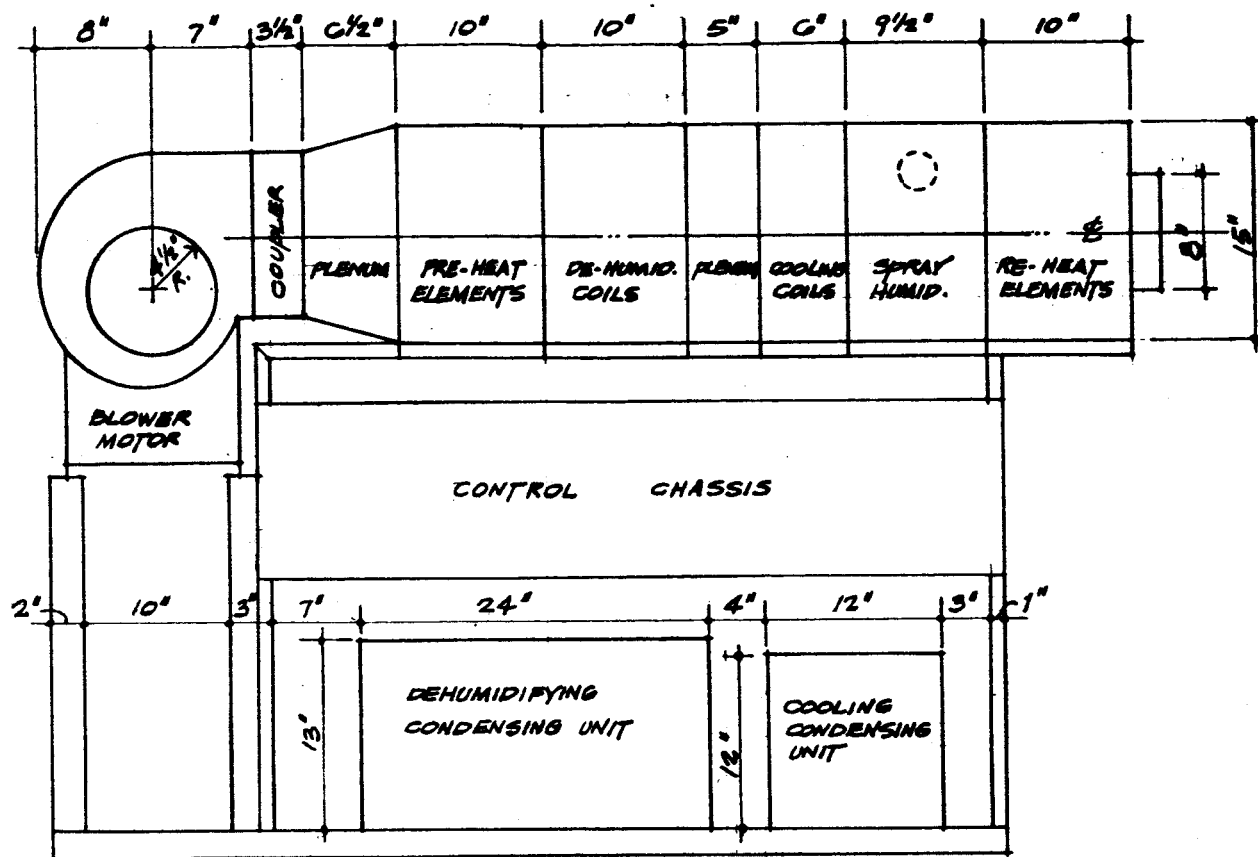
Initial evaluation was interrupted after 48 hours due to the aforementioned icing problems. Modifications to eliminate this and other design deficiencies are now being completed and a comprehensive evaluation of the performance potential of the entire unit will soon be made; however, the data available from the first limited evaluations indicated the following with relation to the Air Handling Chassis: a) the unit would meet the original temperature specification of  $10^{\circ}$ - $35^{\circ}$  centigrade, b) the lower humidity specifications would not be met. The unit consistently delivered a low relative humidity of 52% as contrasted to the desired low of 10%; however, this was as low a figure as could be obtained with the dehumidifying system supplied. Improvement of this figure could be obtained with a more complicated and expensive dehumidifying system.

#### SUMMARY

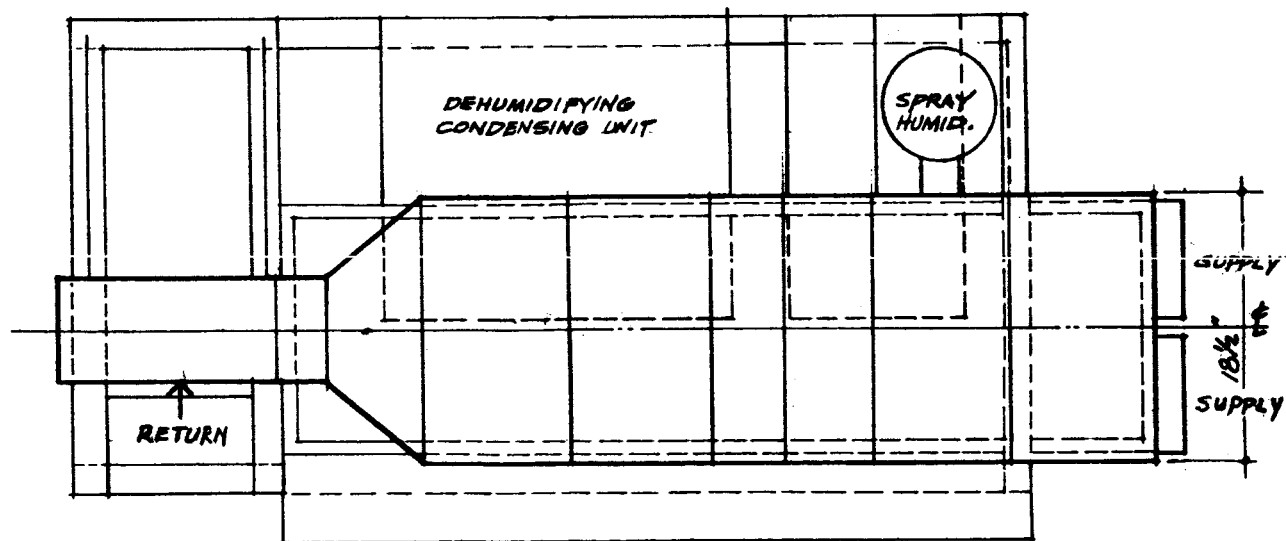
In spite of the use of funds other than those supplied by the National Science Foundation, budget considerations did not allow realization of all of the original design specifications. The chamber described above, however,

not only represents a considerable advancement over modified animal tents which have been used as plant growth chambers but is also better in all respects than any of the germfree plant growth chambers described in the literature and reviewed in the original proposal. "Second generation" growth chambers are now being designed using the prototype for testing features likely to be incorporated.

In summation, the prototype represents a significant step toward achieving the goal of a practical environmental chamber for gnotobiotic growth of plants.



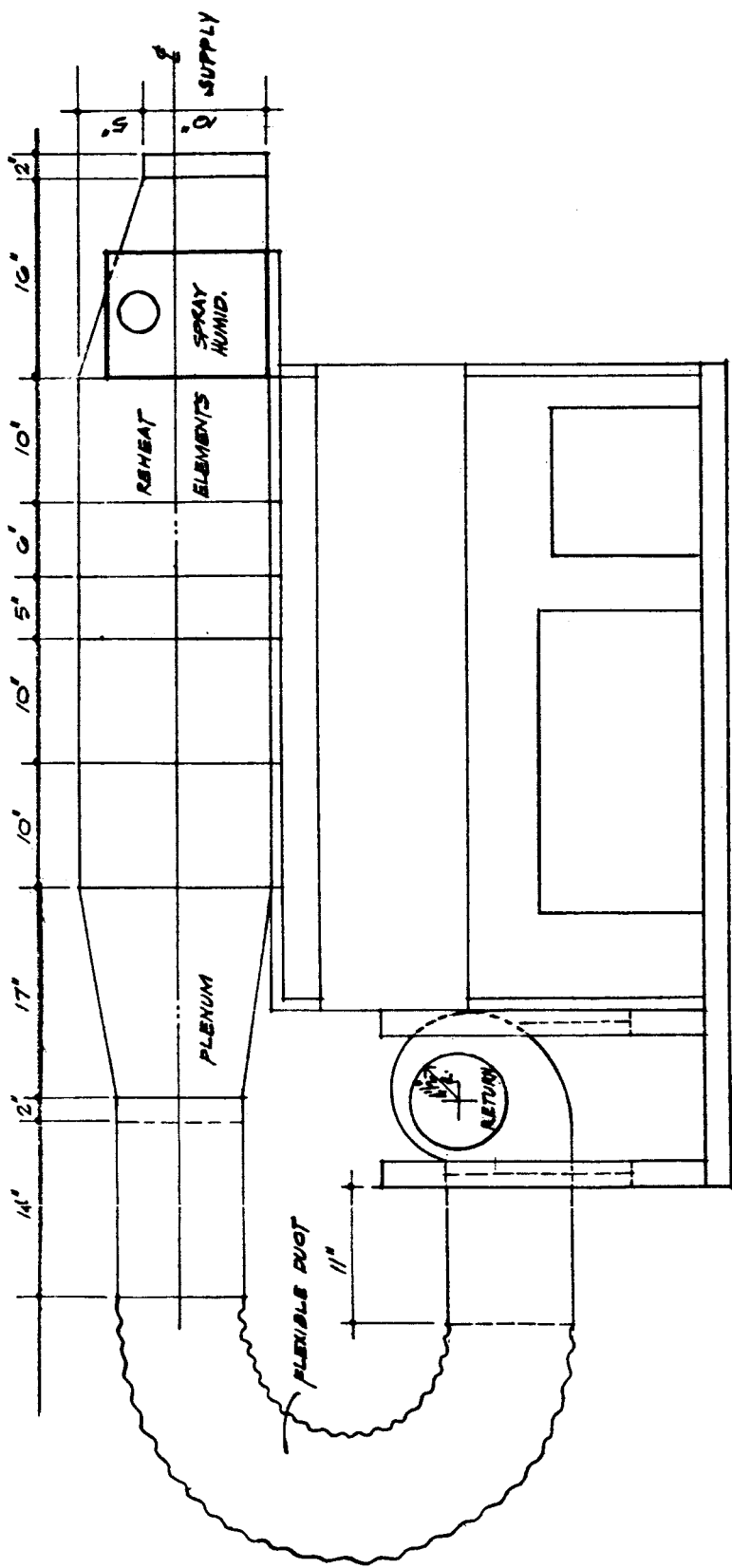
SIDE VIEW



TOP VIEW

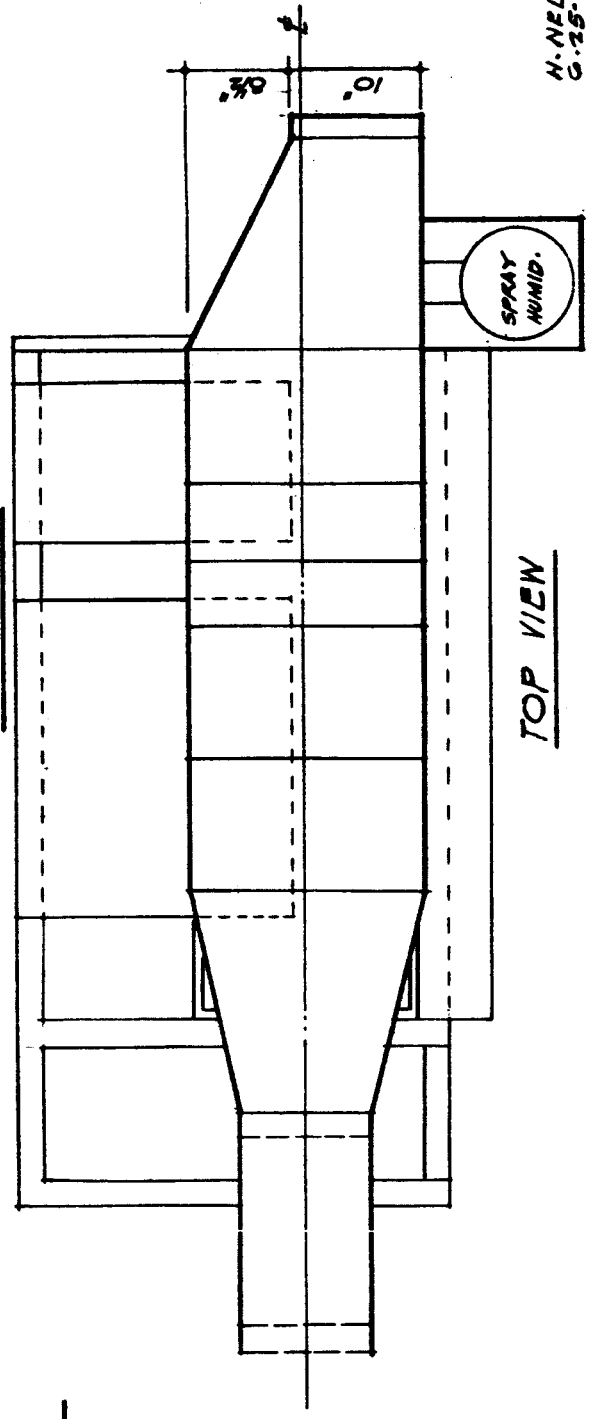
ORIGINAL CHASSIS DRAWING 1-A  
SCALE 1/4" = 1'-0"

H. NELSON  
6-25-66



SIDE VIEW

DRAWING 1B  
SCALE 1"=1'-0"



TOP VIEW

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C. 15-66





DRAWING 2A-2B ORIGINAL CHAMBER & MODS.  
SCALE 1 1/2" = 1'-0"

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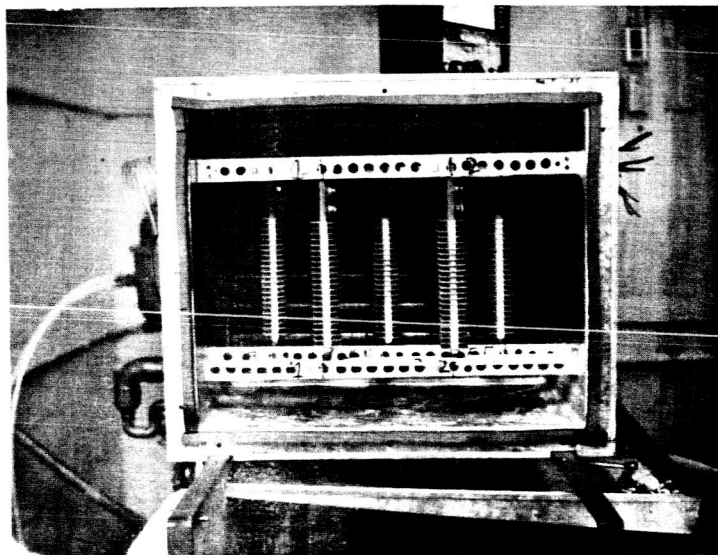
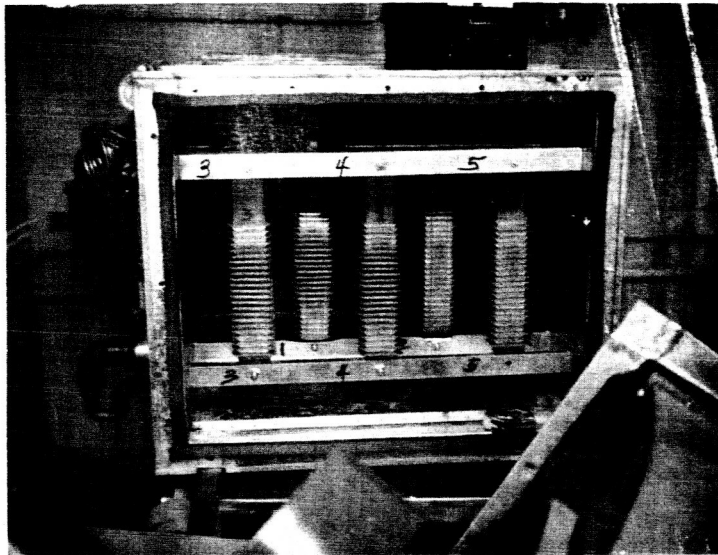


Figure 3

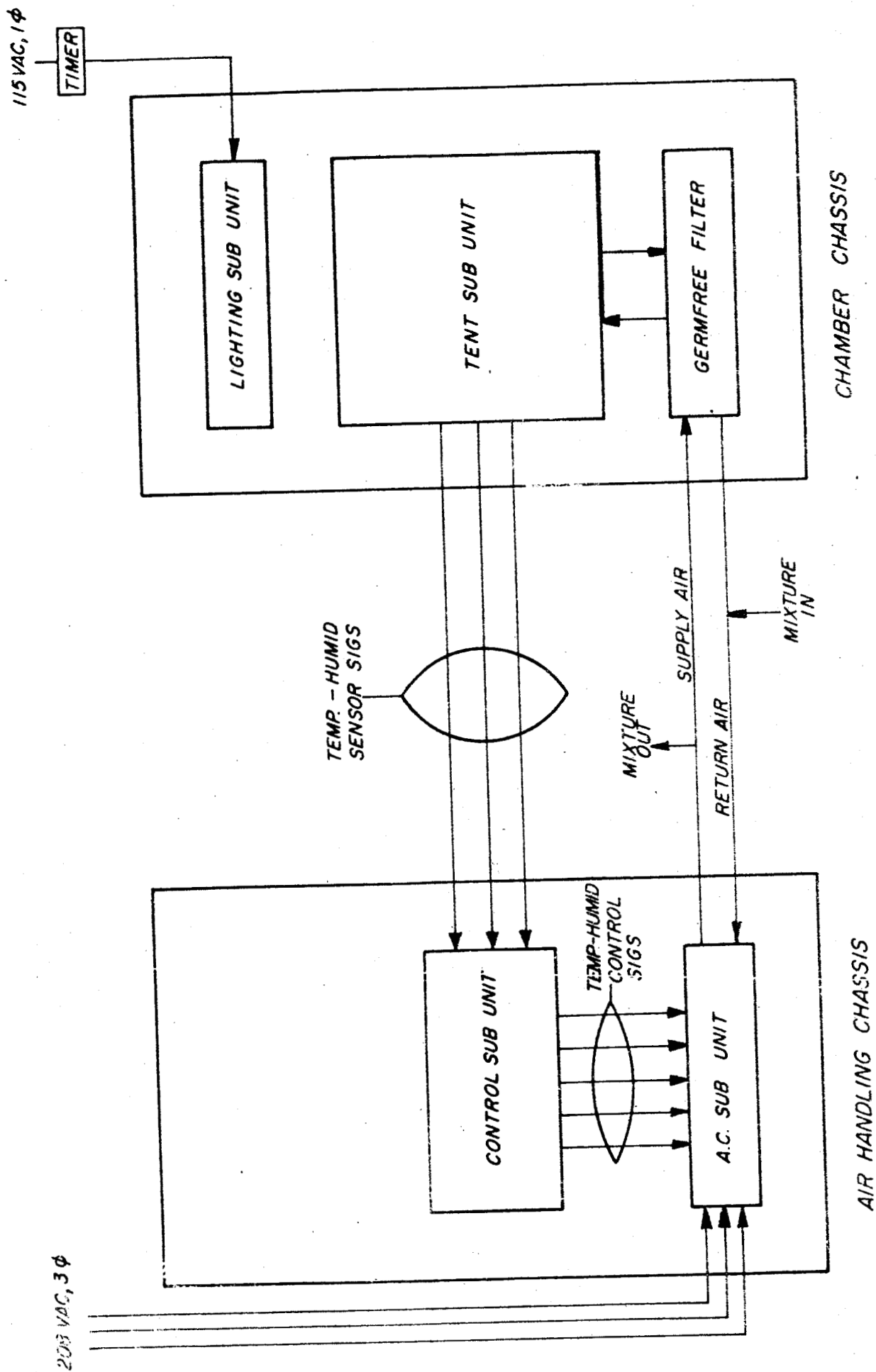


FIG. 4 SYSTEM BLOCK DIAGRAM